Purchased by the USDA Forest Service for official use

Root Stripping of Ectomycorrhizae Decreases Field Performance of Loblolly and Longleaf Pine Seedlings

Donald H. Marx *and* **Glyndon E. Hatchell,** *Institute for Mycorrhizal Research and Development, Forestry Sciences Laboratory, Athens, GA.*

ABSTRACT. Survival and growth of loblolly and longleaf pine seedlings were significantly reduced after 2 years in the field by physical removal (stripping) of specific ectomycorrhizae before planting. The tests were done in South Carolina during the drought years of 1981 and 1982. Loblolly pine seedlings were all the same size at planting. Two sizes of longleaf pine seedlings were produced by varying nursery bed density. After 2 years, mean survival for loblolly pine was 71, 60, 56 and 39% for seedlings with Pisolithus tinctorius *(Pt) ectomycorrhizae that had 0, 33, 66, and 100%, respectively, of the ectomycorrhizae stripped. Pt seedlings with 0 or 33% stripping of ectomycorrhizae were significantly larger than seedlings with more Pt ectomycorrhizae removed. Mean survival of loblolly pine with ectomycorrhizae from natural inoculation (NI) was 61, 52, 39, and 38% with 0, 33, 66, and 100%, respectively, of the ectomycorrhizae stripped. Growth of NI seedlings was significantly less than that of Pt seedlings; NI seedlings also produced fewer new lateral roots than Pt seedlings in the first growing season. Survival and early growth (stem caliper increment) of longleaf pine seedlings were affected more by stripping ectomycorrhizae than by different initial root-collar diameters caused by different bed densities (7.5 and 15 seedlings per ft²). Survival of seedlings from the low bed density averaged (Pt and NI) 59, 48, and 37%, and that of seedlings from the higher bed density averaged 63, 40, and 27% after stripping 0, 50, and 100% of the ectomycorrhizae, respectively. Seedlings from the higher bed density with Pt ectomycorrhizae survived better (48%) than those with NI ectomycorrhize (38%). Thus, the quality and quantity of the ectomycorrhizal condition of loblolly and longleaf pine seedlings from the nursery are highly significant in determining outplanting success, especially during years of low rainfall.*

South. J. Appl. For. 10:173–179, Aug. 1986

At the Southeastern Station's Institute for Mycorrhizal Research and Development, much of our effort during the last several years has been concentrated on the establishment of a single fungal species, *Pisolithus tinctorius* (Pers.) Coker and Couch (Pt) on the roots of conifer seedlings. Recently, a commercial formulation of Pt vegetative inoculum labeled MycoRhiz® (Marx et al. 1982, 1984) and a machine to apply the inoculum to bareroot seedling nursery beds (Cordell et al. 1981) were developed. During tests of the inoculum and equipment we observed that many ectomycorrhizae were being stripped from the lateral roots of seedlings by certain seedling harvesters. Seedlings that were carefully lifted by hand lost fewer ectomycorrhizae.

We suspected that the loss of these ectomycorrhizae during seedling lifting was detrimental to their field performance, and the two studies described here were done to determine how detrimental. In the first study, different amounts of ectomycorrhizae formed by Pt or native nursery fungi on loblolly pine (*Pinus taeda* L.) roots were mechanically removed, and the seedlings were outplanted. In the second study, similar treatments were applied to longleaf pine (*P. palustris* Mill.) seedlings of two sizes. In both studies, field performance was monitored for two years. In the first study, lateral root regeneration was one of the variables observed.

MATERIALS AND METHODS

This section describes the details of our studies. They are, necessarily, somewhat complex and need not be completely understood to grasp the importance of the results. We present these details primarily to illustrate that the methods were based on standardized proven research procedures.

Loblolly Pine

Seedling production. Improved (state seed orchard sources) loblolly pine seedlings were grown in the Taylor State Nursery, Trenton, SC, in a section that grew cover crops (sorghum and rye) in 1979. Rye was turned under in June 1979. Soil was fumigated with 392 kg/ha (350 lb/ac) of Dowfume® MC-33 in October 1979. In March 1980, 0-9-27 fertilizer was applied at 560 kg/ha (500 lb/ac). Biological assay (Marx et al. 1984) of soil samples collected in April 1980 indicated that fumigation effectively eliminated pythiaceous fungi and plant parasitic nematodes. In mid-April 1980, MycoRhiz® was applied to soil in 22.9 m (25 ft) long plots in each of 5 blocks by machine injecting approximately 33 ml (1 oz) MycoRhiz®/ft² in 8 seed drill rows. In each row a band of inoculum 5 cm (2 in.) wide was placed 5 cm deep in soil. MycoRhitz® was a 1980 formulation (batches 1, 2, and 3) of Pt isolate 250 that was also used in other nursery studies (Marx and others 1984). Seeds treated with Arasan® and latex sticker were sown and mulched 1.3 cm (0.5 in.) deep with nonfu-

migated pine straw. Naturally inoculated (control) seedlings from the same seed source were grown in adjacent, replicated, noninoculated plots. Soil in these plots was mechanically disrupted by machine to simulate the disturbance of inoculation. Soil nutrient content at study installation was 215, 77, 49, 19 and 235 μg/g of total N, available P (Bray II), and exchangeable K, Mg, and Ca, respectively.[1] Organic matter content was 1.72%, and pH was 5.6.

During the growing season, all seedlings received two applications of NH_4NO_3 each at 34 kg N/ha (30 lb N/ac), K_2O at 34 kg/ha (30 lb/ac), 17 sprays of Fermate® each at 1.7 kg/ha (1.5 lb/ac), 3 applications of Goal® each at 1.16 liter/ha (2 pt/ac), one application each of TOK at 9.28 liter/ha (1 gal/ac), Captan® at 1.68 kg/ha (1.5 lb/ac), and Sevin® at 1.4 kg/ha (1.25 lb/ac). Seedlings were top-pruned at 20 cm (8 in.) in mid-August and September.

Seedling treatments. In mid-December 1980, seedlings were undercut and carefully hand-lifted from three 0.55 × 1.1 m (2 × 4 ft) subplots in each of the 5 Pt-inoculated and the 5 naturally inoculated (NI) plots. From each ectomycorrhizal treatment (Pt or NI) 1,480 seedlings of similar height, root-collar diameter, and total ectomycorrhizal development were selected and then randomly divided into 4 lots of 370 seedlings each. Variation in parameters was ±5%. These 4 lots of seedlings/ectomycorrhizae treatment were stripped by hand of 100, 66, 33, or 0% (intact) of short roots and ectomycorrhizae. Stripping of short roots and ectomycorrhizae was accomplished by pulling a wet, individual root system through a loose fist 4, 2, or 1 time to produce seedlings with approximately 100, 66, or 33% feeder root stripping, respectively. Seedlings were kept moist during these procedures and were exposed no longer than 15 sec. After the root stripping treatment, each of the 8 seedling treatments (Pt and NI each with 100, 66, 33, and 0% root stripping) were further divided into 5 blocks (field replicates) of 74 seedlings each. Ten of these seedlings were selected at random and measured for height, root-collar diameter, top and root fresh weights, percent short roots ectomycorrhizal with specific fungi, and number of first-order lateral roots. The remaining 64 seedlings in each treatment/replicate were bundled, packed in moist peat moss, and stored at 5°C (41°F) in plastic-lined, kraft paper seedling bags for 11 days before outplanting.

Outplanting. Seedlings were outplanted in Compartment 11 on the Savannah River Forest Station, Aiken, SC. A mature stand of loblolly pine was clearcut in 1979, and the site was burned, stump-sheared, root-raked of most slash, and windrowed in December 1980. Five blocks each 229 × 18 m (750 × 60 ft) were established. Each block was divided into eight 18 × 18 m (60 × 60 ft) plots; each plot was separated by a 9 m (30 ft) border on all sides. In early January 1981, 49 permanent plot seedlings were hand-planted on 3.05 m (10 ft) centers. Each plot contained 7 rows of 7 seedlings. The remaining 15 seedlings/treatment/plot were planted in randomly selected rows of permanent plot seedlings for later removal and destructive root assays. Soil moisture was approximately 25% of field capacity at planting time. Soil fertility did not vary by block (8 random samples at 0- to 8-inch depth/block) and averaged 250, 6, 17, 10, and 93 μg/g of total N, available P, and exchangeable K, Mg, and Ca, respectively. Organic matter was 1.61%, and pH was 4.7. The soil series is Orangeburg.

Data collection and analysis. In mid-March, May, and July 1981, 3 or 4 interplanted seedlings/plot were randomly selected and carefully excavated by shovel. The number of seedlings excavated at each time varied because of the variation in survival among the initial 15 interplanted seedlings/plot. The root/soil mass was carefully rinsed in water, and the roots were carefully separated. All white lateral root tips were counted, and lengths of all lateral roots were measured. Ectomycorrhizae were visually assessed on the entire root system on site without magnification. Survival of permanent plot trees was also determined at each sampling period. In January 1982 and 1983, height and root-collar diameters of permanent plot trees were measured. These data were used to compute seedling and plot volume indices, which are experimentally proven, nondestructive surrogate measures of total aboveground dry weights of southern pine seedlings (Marx et al. 1977, Ruehle et al. 1984, Hatchell et al. 1985). Data were subjected to analysis of variance, and significant treatment means were further separated with Duncan's Multiple Range Test at $P = 0.05$.

Longleaf Pine

Seedling production. Longleaf pine seedlings (North Florida seed source) were grown at the Whitehall Experimental Tree Nursery, Athens, GA. Four wood-framed beds each 1.2 × 6.1 × 0.6 m deep (4 × 20 × 2 ft) were constructed on level ground and filled to a depth of 0.3 m (1 ft) with crushed granite. A 2:1:1 volume mixture of forest clay loam:sand:milled pine bark was used to fill the remaining bed volumes. Soil fertility was 330, 14, 68, 30, and 189 μg/g of total N, available P, and exchangeable K, Mg, and Ca, respectively. Organic matter content was 2.32%, and pH was 5.2. Soil mixture was fumigated at recommended rates with Dowfume® MC-2 under clear polyethylene plastic, which was removed after 5 days. In mid-April 1980, leached and dried vegetative inoculum of Pt (isolate 250), prepared by the methods of Marx et al. (1984), was broadcast at the rate of 0.54 liter/m^2 (1.7 oz/ft^2) of soil surface on two beds and mixed by hand tools into the upper 10 cm (4 in.) of soil. Inoculum had a bulk density of

[1] Chemical analyses of all soil samples reported here were done by A&L Agricultural Laboratories, 411 N. Third St., Memphis, TN.

358 g/liter, a moisture content of 27%, and a pH of 4.5. Fertilizer (10-10-10) was applied at the rate of 560 kg/ha (500 lb/ac) at the same time. Fertilizer was applied at the same rate and mixed in soil of the remaining two beds, which were not artificially inoculated; these were maintained to produce seedlings with naturally occurring ectomycorrhizae (NI). Longleaf pine seed were sown on the 4 beds by hand in rows 2.4 cm (6 in.) apart. Before sowing, seeds were dewinged by hand and treated with Arasan® and latex. After sowing, Captan® at 2.8 kg ai/ha (2.5 lb ai/ac) was applied as a drench to each bed. In early June, seedlings in a Pt and an NI bed were thinned to a density of 80 seedlings/m^2 (7.5/ft^2); the remaining Pt and NI beds were thinned to 160 seedlings/m^2 (15/ft^2). In mid-June, July, August, and September, 56 kg N/ha (50 lb N/ac) or NH_4NO_3/application were applied to each bed. In late August and September, 28 kg K/ha (25 lb K/ac) of KCl/application were also applied to each bed.

In early February 1981, seedlings were vertically root-pruned between rows with shovels, undercut to a depth of 20 cm (8 in.), hand-lifted and graded. Diseased seedlings or those with root-collar diameters less than 9 mm (approximately ⅓ in.) were discarded as culls. Fifteen bundles of 25 plantable-size seedlings from each of the four nursery treatments (Pt and NI in two bed densities) were prepared. Each nursery treatment was then randomly divided into three groups of seedlings of five bundles each. Seedlings in one group had all short roots and ectomycorrhizae stripped (100%), another group was half stripped (50%) throughout their root length, and the last group was left intact (0%). Short roots and ectomycorrhizae were stripped by hand using the same procedure as for the loblolly pine seedlings. Seedlings were rinsed in water, visually assessed without magnification for ectomycorrhizal development, and measured for root-collar diameter. Within treatments, these parameters did not vary by more than ±7%. Seedlings were packed in moist peat moss in plastic-lined kraft paper bags and stored at 5°C overnight before outplanting.

Outplanting. The outplanting site is on the Savannah River Forest Station, Aiken, SC. A mature stand of loblolly pine was clearcut in 1979, and the site was prepared for planting by K-G blading, root-raking into windrows, and disking. The soil series is Blanton. Soil fertility did not vary significantly by block and averaged 404, 6, 17, 8, and 70 μg/g of total N, available P, and exchangeable K, Mg, and Ca, respectively. Organic matter content was 1.63%, and pH was 4.6. The upper 1 to 2 m (40 to 80 in.) of soil depth consist of sand and loamy sand. Five blocks, each 30 × 23 m (100 × 75 ft), were laid out. Each block was divided into two 12 × 23 m (40 × 75 ft) plots. All blocks and plots within blocks had 6 m (20 ft) isolation strips separating them. One randomly selected plot was hand-planted to single rows of 25 seedlings for each of the 6 treatments of Pt; the other plot was hand-planted to 6 rows of appropriate NI seedlings. Row spacing was 2.4 m (8 ft) and within-row spacing was 0.9 m (3 ft). Seedlings were planted in early February 1981, when soil moisture was about 50% of field capacity. The experimental design was a split-plot (Pt or NI). Each half-plot contained 6 treatment rows for all combinations of 2 bed densities and 100, 50, and 0% stripping of ectomycorrhizae.

Data collection and analysis. Survival was determined after three months, one, and two years. Three seedlings/treatment/replicate were excavated and assessed for ectomycorrhizal development after the first year. It was assumed that these seedlings would have survived to the end of the second year had they not been removed. Root-collar diameters were obtained after one month (initial) and at the end of each growing season. Survival and diameter data were statistically analyzed as in the loblolly pine study. After two years in the field less than 12% of the seedlings had initiated height growth, and the pattern of growth initiation was not related to treatment. Height data, therefore, are not reported here.

RESULTS

Loblolly Pine

Height, root collar, and fresh weight of seedlings prior to outplanting were not affected by the ectomycorrhizal type or the root-stripping treatments. Thus, grading of seedlings to standardize their initial size was successful, and removal of ectomycorrhizae apparently did not significantly change T/R ratios (fresh weight of tops/roots) or number of lateral roots. At planting time seedling heights were 28.8 cm (11.3 in.), root-collar diameters were 0.52 cm (0.2 in.), top weights were 13.4 g (0.5 oz), root weights were 4.9 g (0.17 oz), number of first-order lateral roots was 8. The only seedling measurements different at outplanting were the amounts of Pt and NI ectomycorrhizae remaining on the root system. Total ectomycorrhizal development following 0, 33, 66 and 100% stripping of Pt averaged 44, 27, 14 and 4%, and the Pt indices were 76, 63, 54, and 47, respectively. Pt ectomycorrhizae represented about 86% of the total ectomycorrhizae on Pt seedlings; other ectomycorrhizae were formed by *Thelephora terrestris* (Tt) Ehrh. ex Fr. Total ectomycorrhizal development on the NI seedlings after stripping averaged 45, 31, 16, and 2% as stripping intensity increased. The NI ectomycorrhizae were formed primarily by *T. terrestris*. Although values are not presented, the number of nonmycorrhizal short roots on the lateral roots was approximately the same regardless of the amounts of ectomycorrhizae stripped from the root systems.

Rainfall during 1981 and 1982 on the study site was unusually low. Only 2.5 mm (0.1 in.) of rainfall were recorded in a rain gauge

located about 3 km (1.8 mi) from the test area for the first 4 months after seedling planting. An additional 71 mm (2.8 in.) were recorded during the next 4 months. Nearly normal rainfall amounts of 58 mm/mo (2.3 in/mo) occurred from August to December. The second growing season was as droughty as the first. Only 904 mm (35.6 in.) of rainfall fell near the study area—about 356 mm (14 in.) less than normal.

Periodic examination of seedlings revealed that the development of new lateral roots and their total lengths were related to both the original ectomycorrhizal condition (Pt or NI) and the degree of removal of the ectomycorrhizae (Table 1). The relative number of first-order lateral roots (7 to 9/seedling) did not significantly change during the first 6 months. The development of new lateral roots was apparently correlated with increased rainfall during the 4th through 6th months. Generally, seedlings with more Pt ectomycorrhizae at planting (0 and 33% stripped) had more new lateral roots and greater total length of lateral roots at each assessment period than did seedlings with NI ectomycorrhizae or those with more Pt ectomycorrhizae stripped.

Ectomycorrhizal development on the original and newly formed lateral roots was correlated with the ectomycorrhizal treatment in the nursery. Development of ectomycorrhizae by Pt on lateral roots was greatest on seedlings with the most Pt ectomycorrhizae (0 and 33% stripped) at planting. Seedlings that had 66 and 100% of their ectomycorrhizae stripped formed more ectomycorrhizae from resident fungi on the outplanting site than other treatments. These ectomycorrhizae were not morphologically similar to either Pt ot Tt ectomycorrhizae. After six months, there were few differences in total ectomycorrhizal development on seedlings regardless of initial treatment.

Seedling survival was significantly affected by root stripping of Pt and NI ectomycorrhizae (Table 2). The drought during the first six months of this study probably accounted for the high mortality, which was greatest between the 4th and 6th months. However, seedlings with the greatest amount of residual ectomycorrhizae at planting survived better than those with lesser amounts. Seedlings with Pt ectomycorrhizae generally survived better than NI seedlings, and seedlings with 100% of their ectomycorrhizae stripped survived poorly regardless of the specific ectomycorrhizal type at planting.

Seedling growth was also significantly affected by the treatments. Unfortunately, over 97% of the seedlings in this study were damaged by tip moths during each

Table 1. Lateral root and ectomycorrhizal development of loblolly pine seedlings after two, four, and six months in the field with different percentages of specific ectomycorrhizae stripped from lateral roots before field-planting.[a]

Ectomycorrhizal condition[b]	Percent removal	No. new lateral roots[c]	Total length of new lateral roots[c]	% Ectomycorrhizae[d] with:	
				Pt	All fungi
		After 2 months in field			
Pt	0	5.2a	5.6a	22a	34a
NI		2.5b	3.7a	0	29ab
Pt	33	4.0a	4.3a	18a	23b
NI		1.3b	2.4b	0	21b
Pt	66	3.6ab	4.9a	6b	13c
NI		0.3c	1.5b	0	10c
Pt	100	2.3b	3.2a	1c	3c
NI		1.1bc	1.6b	0	2c
		After 4 months in field			
Pt	0	8.6a	14.4ab	34a	41a
NI		6.0ab	12.8b	0	44a
Pt	33	9.8a	17.9a	28ab	36a
NI		4.0c	9.9c	0	38a
Pt	66	7.8ab	13.8ab	9b	17b
NI		2.8c	10.3bc	0	17b
Pt	100	6.4ab	12.1b	3b	14b
NI		2.9c	5.3c	0	18b
		After 6 months in field			
Pt	0	16.6a	112.1a	39a	53a
NI		15.7a	77.3b	0	39ab
Pt	33	17.8a	99.3a	31a	48ab
NI		9.0b	63.3bc	0	51a
Pt	66	13.8ab	70.4b	12b	44ab
NI		7.8b	35.8c	0	31b
Pt	100	13.4ab	81.1ab	3c	39ab
		8.9b	49.8c	0	41ab

[a] Within columns means sharing common letters in the same month of assessment are not significantly different at $P = 0.05$.
[b] Pt = *Pisolithus tinctorius;* NI = naturally inoculated in nursery.
[c] Only new lateral roots of all orders recorded. At planting all seedlings had initially 7 to 9 first-order lateral roots.
[d] Ectomycorrhizae assessed on initial and new lateral roots.

Table 2. Survival over two years of loblolly pine seedlings with different amounts of specific ectomycorrhizal stripped before outplanting.[a]

Ectomycorrhizal condition	Stripping	Survival after				
		2 mo	4 mo	6 mo	12 mo	24 mo
		(%)				
Pt	0	97a	95a	76a	73a	72a
NI		93a	91a	68a	64ab	60b
Pt	33	94a	93a	68a	66a	60b
NI		94a	93a	55ab	52b	52bc
Pt	66	93a	90a	65a	62a	55bc
NI		85b	82b	52ab	47bc	38c
Pt	100	90ab	86ab	48b	43bc	39c
NI		83b	82b	47b	39c	37c

[a] Within columns means sharing a common letter are not significantly different ($P = 0.05$).

growing season. Based on the length of dead terminals, this infestation caused an average loss of about 18% of new height growth each year. The incidence and severity of damage were not related to treatment.

Height growth of seedlings with Pt ectomycorrhizae was not affected by stripping of ectomycorrhizae (Table 3). Effects may have been marked by severe tip moth infestations. However, seedlings with 0 or 33% stripping of Pt ectomycorrhizae had significantly larger root-collar diameters and individual volumes than did seedlings with greater amounts of Pt ectomycorrhizae removed. Removal of ectomycorrhizae from NI seedlings did not affect root-collar diameters, but NI seedlings with all ectomycorrhizae stripped had significantly less height growth than those with intact root systems. NI seedlings with 0 or 33% root stripping of ectomycorrhizae generally had greater individual volumes than those with more of their ectomycorrhizae stripped. Also, seedlings with 0 or 33% root stripping of Pt ectomycorrhizae were generally larger than NI seedlings with the same stripping treatment. Plots of Pt seedlings with 0, 33, or 66% root stripping of ectomycorrhizae had significantly greater PVI than those of NI seedlings with similar amounts of natural ectomycorrhizae. PVI of seedlings of either ectomycorrhizal type decreased in proportion to the percentage of ectomycorrhizae removed. However, PVI were not different for NI seedlings with 66 or 100% root stripping of ectomycorrhizae and Pt seedlings with 100% stripping of ectomycorrhizae.

Longleaf Pine

The root-stripping treatments did not significantly affect any of the initial measurements on the longleaf pine seedlings (Table 4). Seedling density in the nursery bed had a significant effect on root-collar diameter and degree of Pt ectomycorrhizal development. Seedlings grown at 7.5/ft² bed density were significantly larger but had fewer Pt ectomycorrhizae than seedlings grown at 15/ft². Pt indices averaged 78 before stripping. There were no differences in root-collar diameters of seedlings between any of the ectomycorrhizal conditions at the same bed density. None of the nursery treatments affected the number of first-order lateral roots on longleaf pine seedlings.

Assessments of ectomycorrhizae on seedlings after the first growing season showed that the seedlings with the greatest percentage of ectomycorrhizae at planting had the greatest amounts (36 to 48%) after 1 year. Both Pt and NI seedlings with 100% of the ectomycorrhizae stripped had only about half (28 to 34%) this amount after 1 year. The deep penetrating and meandering root habit of longleaf pine seedlings in this sandy soil made it difficult to locate lateral roots and make accurate assessments of the ectomycorrhizae. Excavations after the first year did indicate, however, that the Pt ectomycorrhizae formed in the nursery persisted well and spread to new lateral roots on all seedlings except for those with all ectomycorrhizae stripped before planting. These latter seedlings formed morphologically different ectomycorrhizae apparently from site-resident fungi. The relative number of first-order lateral roots (10 to 14/seedling) did not change after 1 year in the field.

Longleaf pine seedling mortality was high during the first year (1981) especially during the early summer months with low rainfall. Trends in survival through the end of the second year followed those of loblolly pine discussed earlier. Only second-year survival data is presented for longleaf pine seedlings (Table 5). Removal of different amounts of ectomycorrhizae before planting had a greater effect on seedling survival than either the specific ectomycorrhizal type or seedling size as affected by bed density. Generally, the smaller seedlings from the higher bed density did not survive as well as

Table 3. Growth of loblolly pine seedlings after two years with 0, 33, 66, or 100% root stripping of specific ectomycorrhizae prior to outplanting.[a]

Ectomycorrhizae at planting	Stripping	Height	Root-collar diameter	Seedling volume[b]	PVI × 10²[c]
	(%)	(cm)	(cm)	(cm³)	(cm³)
Pisolithus	0	81.0a	2.16a	490a	166a
tinctorius	33	79.5a	2.10a	451a	133b
(Pt)	66	71.2a	1.79b	324ab	90c
	100	71.5a	1.79b	299b	62c
Natural	0	75.3x	1.91x	364x	108x
(NI)	33	68.5xy	1.86x	312x	79xy
	66	66.9xy	1.82x	282xy	51y
	100	53.2y	1.74x	260y	50y

[a] Within columns means sharing common letters are not significantly different at $P = 0.05$. Means underlined are significantly different from the same ectomycorrhizae stripping treatment with natural ectomycorrhizae.
[b] Seedling volume = (root-collar diameter)² × height.
[c] PVI = plot volume index (seedling volume × number of surviving seedlings per plot).

Table 4. Initial measurements of longleaf pine seedlings from different bed densities after stripping various amounts of specific ectomycorrhizae before outplanting.[a]

Ectomycorrhizal conditions	Bed density	Stripping	Root-collar diameter	No. 1st order lateral roots	Ectomycorrhizae with:	
					Pt	All fungi
	(ft^2)	(%)	(cm)		..... (%)	
	7.5	0	1.18a	11a	36a	50a
		50	1.20a	13a	14b	21b
Pisolithus tinctorius		100	1.13a	10a	1c	3c
(Pt)	15	0	1.00b	12a	58a	65a
		50	0.95b	14a	24b	29b
		100	0.92b	10a	1c	4c
	7.5	0	1.20a	11a	0	22a
		50	1.14a	12a	0	10b
Natural (NI)		100	1.15a	11a	0	3c
	15	0	1.06b	12a	0	32a
		50	1.02b	14a	0	15b
		100	1.03b	11a	0	2c

[a] Within columns means sharing a common letter are not significantly different ($P = 0.05$).

larger seedlings regardless of ectomycorrhizal condition.

Seedling growth, as measured by increase in root-collar diameter (diameter at planting minus second-year diameter), was significantly affected by the three main treatments. The most significant treatment effect was stripping of the ectomycorrhizae (Table 5). Seedlings with all ectomycorrhizae removed had only about half as much increment in root-collar diameter as those with initially intact root systems. Pt seedlings with 50% of the ectomycorrhizae removed grew as much as those with 0% and significantly more than NI seedlings with 50% of the ectomycorrhizae stripped. Generally larger seedlings from the low bed density had significantly more growth than smaller seedlings from the higher bed density.

DISCUSSION

The field performance of loblolly and longleaf pine seedlings was significantly improved by retaining a large percentage of ectomycorrhizae on their lateral roots at planting time. There are strong indications that ectomycorrhizal development by Pt in the nursery affected the physiological condition of the loblolly pine seedlings used in this study. Pt seedlings with all or 66% of the ectomycorrhizae removed before outplanting formed nearly twice as many new second- and third-order lateral roots during the first four months in the field as did NI seedlings with various quantities of ectomycorrhizae stripped. The physiological significance of more roots to seedlings is better survival and growth. The present study clearly indicates that seedlings with the greatest quantity of ectomycorrhizae at planting, especially Pt ectomycorrhizae, are able to rapidly regenerate numerous new lateral roots of greater length, form more new ectomycorrhizae, and, thereby, utilize available water and nutrients from the outplanting site more effectively.

The relative number of first-order lateral roots on loblolly and longleaf pine seedlings in this study remained the same between planting time and the end of the first dormant season. This result confirms the observations of Mexal and Burton (1978) and suggests that this trait is under strong genetic control.

The overall better performance of the larger longleaf pine seedlings verifies earlier reports (Scarbrough and Allen 1954, Derr 1955, Mann 1969). However,

Table 5. Second-year survival and increase in root-collar diameter increments of longleaf pine seedlings from two nursery bed densities after stripping different amounts of specific ectomycorrhizae before outplanting.[a]

Ectomycorrhizal condition	Bed density	Stripping	Survival	Root-collar diameter increment[b]
	(ft^2)	(%)	(%)	(mm)
Pisolithus tinctorius (Pt)	7.5	0	58a	23a
		50	50b	21a
		100	37c	10c
	15	0	68a	18ab
		50	47bc	18ab
		100	28d	6c
Natural	7.5	0	60a	21a
		50	46bc	16b
		100	37c	13bc
	15	0	57a	15ab
		50	32d	13bc
		100	27d	8c

[a] Within columns means sharing a common letter are not significantly different ($P = 0.05$).
[b] Increment = root-collar diameter at planting minus root-collar diameter after two years in the field.

seedling size was not as important in this study as was the ectomycorrhizal condition of the root systems. Survival and growth were as good or better for smaller seedlings (high bed density) with more ectomycorrhizae than larger seedlings (low bed density) with fewer ectomycorrhizae. However, large seedlings with the greatest amounts of ectomycorrhizae survived and grew better than all others, suggesting that both seedling size and quantity of ectomycorrhizae are highly important determinants of good quality in longleaf pine seedlings.

These results confirm the findings of others (Wakeley 1954, Stone et al. 1963, Tanaka et al. 1976, Mexal and Burton 1978, Barnard et al. 1981, Tinus 1981, Brissette and Lantz 1982, Burdett et al. 1983) that quality of pine seedlings cannot be solely based on measurements such as height, root-collar diameter, seedling weight, or T/R ratios. In this study, removal of all ectomycorrhizae on Pt or NI seedlings did not significantly affect root weights, T/R ratio, or other quantitative measurements, but did markedly affect their field performance. Every effort should be made in nurseries to manage for abundant ectomycorrhizae on seedlings by use of appropriate cultural practices or by artificial inoculation, and to assure that abundant ectomycorrhizae are retained on seedlings during the lifting, packing, storage, and field planting operations for maximum field performance. □

Literature Cited

BARNARD, E. L., C. A. HOLLIS, AND W. L. PRITCHETT. 1981. A comparative evaluation of seedling quality in commercial forest nurseries in Florida. P. 34–41 *in* Proc. 1980 South. Nursery Conf., USDA For. Serv. Tech. Publ. SA-TP17.

BRISSETTE, J. C., AND C. W. LANTZ. 1982. Seedling quality: summary of a workshop. P. 303–305 *in* 1982 Proc. South. Nursery Conf. East. Sess. Ga. For. Comm.

BURDETT, A. N., D. G. SIMPSON, AND C. F. THOMPSON. 1983. Root development and plantation establishment success. Plant & Soil 71:103–110.

CORDELL, C. E., D. H. MARX, J. R. LOTT, AND D. S. KENNEY. 1981. The practical application of *Pisolithus tinctorius* ectomycorrhizal inoculum in forest tree nurseries. P. 38–42 *in* Forest regeneration. Am. Soc. Agric. Eng.

DERR, H. J. 1955. Seedbed density affects longleaf pine survival and growth. Tree Planter's Notes 20:28–29.

HATCHELL, G. E., C. R. BERRY, AND H. D. MUSE. 1985. Nondestructive indices to measure growth of young southern pines on research plots. For. Sci. 31:417–425.

MANN, W. J., JR., 1969. At last—longleaf can be planted successfully. For. Farmer 18:6–7, 18–19.

MARX, D. H., W. C. BRYAN, AND C. E. CORDELL. 1977. Survival and growth of pine seedlings with *Pisolithus* ectomycorrhizae after two years on reforestation sites in North Carolina and Florida. For. Sci. 23:363–373.

MARX, D. H., ET AL. 1982. Commercial vegetative inoculum of *Pisolithus tinctorius* and inoculation techniques for development of ectomycorrhizae on container-grown tree seedlings. For. Sci. 28:373–400.

MARX, D. H., ET AL. 1984. Commercial vegetative inoculum of *Pisolithus tinctorius* and inoculation techniques for development of ectomycorrhizae on bare-root tree seedlings. For. Sci. Monogr. 25. 101 p.

MEXAL, J. G., AND S. BURTON. 1978. Root development of planted loblolly pine seedlings. P. 85–90 *in* Proc. root form of planted trees symp., E. Van Eerden and J. M. Kinghorn, eds. B.C. Minist. For. & Can. For. Serv. Jt. Rep. 8. 357 p.

RUEHLE, J. L., D. H. MARX, AND H. D. MUSE. 1984. Calculated nondestructive indices of growth response for young pine seedlings. For. Sci. 30:467–72.

SCARBROUGH, N. M., AND R. M. ALLEN. 1954. Better longleaf seedlings from low-density nursery beds. Tree Planter's Notes 18:29–32.

STONE, E. C., ET AL. 1963. Variation in the root regeneration potentials of ponderosa pine from four California nurseries. For. Sci. 9:217–25.

TANAKA, Y., J. D. WALSTAD, AND J. E. BORRECCO. 1976. The effect of wrenching on morphology and field performance of Douglas-fir and loblolly pine seedlings. Can. J. For. Res. 6:453–58.

TINUS, R. W. 1981. Concepts for seedling production and handling being developed in New Zealand. P. 56–60 *in* Forest regeneration. Am. Soc. Agric. Eng.

WAKELEY, P. C. 1954. Planting the southern pines. USDA For. Serv. Agric. Monogr. 18. Wash., D.C. 233 p.